

Abnormal Growth Factor/Cytokine Network in Gastric Cancer

Eiichi Tahara

Received: 23 December 2007 / Accepted: 18 February 2008 / Published online: 19 March 2008
© The Author(s) 2008

Abstract Gastric cancer cells express a broad spectrum of the growth factor/cytokine receptor systems that organize the complex interaction between cancer cells and stromal cells in tumor microenvironment, which confers cell growth, apoptosis, morphogenesis, angiogenesis, progression and metastasis. However, these abnormal growth factor/cytokine networks differ in the two histological types of gastric cancer. Importantly, activation of nuclear factor- κ B pathway by *Helicobacter pylori* infection may act as a key player for induction of growth factor/cytokine networks in gastritis and pathogenesis of gastric cancer. Better understanding of these events will no doubt provide new approaches for biomarkers of diagnosis and effective therapeutic targeting of gastric cancer.

Keywords Gastric cancer · Growth factor/cytokine · Tumor microenvironment · *Helicobacter pylori* · Inflammation · NF- κ B pathway

Abbreviations

<i>H. pylori</i>	helicobacter <i>pylori</i>
Cag A	cytotoxin-associated gene A
NF- κ B	nuclear factor- κ B
IL	interleukin
TNF	tumor necrosis factor
COX	cyclooxygenase
VEGF	vascular endothelial growth factor

MMP	matrix metalloproteinase
iNOS	inducible nitric oxide synthase
TGF	transforming growth factor
EGF	epidermal growth factor
FGF	fibroblast growth factor

Introduction

Gastric cancer remains the world's second commonest malignancy [1]. However, there is substantial variation in gastric cancer incidence with the highest rates reported from Korea, Japan and Eastern Europe, whereas Western Europe and the US have low incidence rates. The global burden of gastric cancer is shifting rapidly from the developed world to the developing world.

Recent evidence indicates that a large number of genetic and epigenetic alterations in oncogenes, tumor suppressor genes and genetic instability as well as telomerase activation determine a multi-step process of gastric carcinogenesis [1, 2]. However, these molecular events found in gastric cancer differ depending upon the two histological types, intestinal and diffuse types of gastric carcinoma, indicating that intestinal and diffuse type carcinomas have distinct genetic pathway [2]. The evolution of intestinal tumors is generally characterized as progressing through a number of sequential steps including gastritis, intestinal metaplasia, dysplasia and carcinoma, while no preceding steps are detected in the pathogenesis of diffuse carcinoma other than the obvious chronic gastritis.

The most significant advance in the pathogenesis of gastric cancer is involved with the recognition of the role of *H. pylori*

E. Tahara (✉)
Hiroshima University, Hiroshima Cancer Seminar Foundation,
3-8-6 Sendamachi, Naka-ku,
Hiroshima 730-0052, Japan
e-mail: etahara@h-gan.com

as the most important etiological factor for gastric cancer [1]. *H. pylori* infection is associated with chronic inflammation and multistage process of gastric carcinogenesis [3].

Both the complex host/microbial interaction and growth factor/cytokine networks induced by *H. pylori* are mediated by the microenvironment that is an indispensable player in the multistage process of gastric carcinogenesis. This review provides an overview of *H. pylori*-induced inflammation and the molecular and cellular events of tumor microenvironment that underlie gastric cancer.

***H. Pylori*-Induced Inflammation Implicated in Gastric Carcinogenesis**

There are several of *H. pylori*-virulence associated genes such as *cagA*, *vacA*, *iceA* and *babA* [1]. Among them, the *cagA* gene is localized at one end of the *cag* pathogenicity island (PAI) [4]. The *cagPAI* encodes components of a type IV secretion system, by which the *cagA* gene product, CagA, is delivered into gastric epithelial cells [5]. The *cagA*-positive *H. pylori* strains are associated with higher grade of gastric inflammation and higher risk of gastric cancer than the *cagA* negative strains [6]. Prinz et al. [7] reported that CagA+/VacAs1+ strains of *H. pylori* that are blood group antigen-binding adhesion (BabA2) positive are associated with activity and chronicity of gastritis. Adherence of *H. pylori* via BabA2 may play a key role for efficient delivery of VacA and CagA. Moreover, Hatakeyama [8] recently reported that CagA binds an Src homology 2 (SH2) containing tyrosine phosphatase SHP-2 in a tyrosine phosphorylation-dependent manner and activates the phosphatase activity of SHP-2. Deregulation of SHP2 by CagA is an important mechanism by which *cagA* positive *H. pylori* promotes gastric carcinogenesis. Moreover, Hatakeyama [8] found that East-Asian CagA shows stronger SHP2 binding and greater biological activity than Western CagA. Differences in the biological activity of Western and East-Asian CagA proteins, which are determined by variation in the tyrosine phosphorylation sites, may underlie the different incidences of gastric cancer in these two geographic areas.

In addition to deregulation of SHP2 by CagA, *H. pylori* is a potent activating factor of NF- κ B in gastric epithelial cells [9]. Activation of NF- κ B by *H. pylori* infection induces a variety of gene expression including cytokines (IL-1, IL-6, IL-8, TNF alpha), VEGF, COX2, iNOS, cell-cycle regular, MMP2, MMP9 and adhesion molecules [10–12]. Successful eradication of *H. pylori* leads to down regulation of COX2 in the epithelial and stromal cells [2]. High expression of COX2 mRNA, protein, and enzymatic activity is detected in the tumor cells of gastric cancer [2]. Importantly, COX2 activity is induced by a variety of mediators including inflammatory cytokines such as TNF alpha, interferon-gamma and IL-1

[13]. COX2 facilitates tumor growth by inhibiting apoptosis, maintaining cell proliferation and stimulating angiogenesis within cancer cells [14].

H. pylori infection produces reactive oxygen and nitrogen species that cause DNA damage, followed by chronic gastritis and intestinal metaplasia [3]. Nitric oxide generated by iNOS is converted to reactive nitrogen species that bring about direct DNA mutation such as p53 and protein damage, inhibition of apoptosis, and promotion of angiogenesis [15]. Goto et al. [16] reported that the expression of iNOS and nitro tyrosine in the gastric mucosa was significantly high in *H. pylori* infected patients who developed gastric cancer at least two years after the initial biopsies.

Factors Associated with Increased Incidence of Gastric Carcinoma

Three major factors, including environmental factors (diet, obesity, cigarette smoking), host factors (*H. pylori* strains), and genetic factors, cooperatively affect the genesis of gastric cancer [2]. Of these factors, host genetic factors play a critical role in susceptibility to gastric carcinogenesis.

El-Omar [17] reported that pro-inflammatory IL-1 gene cluster polymorphisms increase the risk of gastric cancer and its precursors in the presence of *H. pylori*. Moreover, the pro-inflammatory IL-1 genotypes are associated with an increased risk of both intestinal and diffuse types of gastric cancer [18]. In addition, TNF alpha and IL-10 genotypes are viewed as independent risk factors for gastric cancer [18]. The combination of three or four pro-inflammatory cytokine polymorphisms affecting IL-1 beta (IL-1B-511T), IL-1 receptor antagonist (IL-1RN*2), TNF alpha (TNF-A-308A) and IL-10 (IL-10 haplotype ATA) results in a highly significant increased risk of development of gastric cancer [18]. However, we recently found that IL-10 haplotype (IL-10 GGCG) is associated with increased risk of gastric cancer and high levels of plasma IL-10 serum in atomic bomb survivors [19]. More recently, Ohyauchi et al. [20] reported that IL-8 polymorphism is also associated with increased risk of gastric cancer in the Japanese population. These host genetic factors as well as *H. pylori* strains may determine why some individual infected with *H. pylori* develop gastric cancer while others do not.

Stem Cell Hyperplasia Induced by *H. pylori*

H. pylori infection induces stem cell hyperplasia in chronic inflammation leading to increased mutation and DNA methylation. Stem cells in the gastric epithelium have yet

to be precisely identified, but there are several proposed markers of normal human stem cells in the gastrointestinal tract including Musashi1 (Msi-1), Hes-1 and nuclear beta-catenin [21]. Msi-1 is an RNA binding protein that may be necessary for stem cell maintenance and/or asymmetric cell division. In human gastric epithelium, Msi-1 is expressed at the proliferative regions of the antrum and fundic glands but its expression is decreased in intestinal metaplasia suggesting that Msi-1 is a marker of gastric progenitor cells [22]. Interestingly, *H. pylori* infection strongly induces the expression of Msi-1 that is correlated with *H. pylori* density [23]. Ushijima group [24] recently reported that *H. pylori* infection potently induces methylation of CpG islands (LOX, *HAND1*, *THBD* and *p41ARC*), suggesting that *H. pylori* induces DNA methylation in stem cells. Cell division itself is a promoting factor for *de novo* DNA methylation.

The RNA component of telomerase, hTERC, and the catalytic subunit, hTERT, are essential and the minimum components that are required for telomerase activity [25]. Telomerase activity and TERT expression are necessary for stem cell function [26]. In fact, we found that low levels of telomerase activity exist in Ki-67-positive epithelial stem cells, which simultaneously express hTERT protein and hTERC protein in intestinal metaplasia of the stomach [27]. The levels of hTERT expression and telomerase activity are in parallel with degree of *H. pylori* infection, suggesting that telomerase competence of stem cell hyperplasia caused by *H. pylori* infection may be linked to “chronic mitogenesis” that can facilitate “increased mutagenesis”.

We previously reported that telomerase activity is present in a majority of gastric cancer [2]. The approach for cancer therapy targeting telomerase and/or telomeres has the potential for innovative anti-cancer drugs. Telomestatin, a G-quadruplex stabilizing agent found in natural products, is highly selective for telomere G-quadruplex structures formed by the 3' overhang compared to other compounds [28]. Telomestatin preferentially induces apoptosis in cancer cells or not in normal fibroblasts and epithelial cells [29]. Moreover, Kondo et al. recently reported that loss of heterozygosity and histone hypoacetylation of the PINX1 gene, a possible telomerase inhibitor, are associated with reduced expression in gastric carcinoma [30]. They also revealed that telomerase activity is inhibited with trichostatin (TSA) and nicotinamide (NAM) in gastric cancer cells even if hTERT expression is not changed. These results provide a possibility for cancer therapy with HAM [30].

Currently, a body of evidence indicates that bone marrow-derived stem cells can engraft and differentiate into nonhematopoietic cells of ectodermal, mesodermal and endodermal tissues other than hematopoietic tissues. These stem cells also contribute to cancer stroma in mouse models [21]. Recently, Taketo group [31] found that SMAD4-deficient mouse colorectal cancer cells produce CC chemo-

kine ligand 9 (CCL9) and recruit the MMP-expressing immature myeloid cells (iMC) that express CC chemokine receptor 1 (CCR1) and that lack of CCR1 prevents the accumulation of MMP-expressing iMC at the invasion front and suppresses tumor invasion. These exciting results suggest that human gastric cancers also may have the possibility to exhibit interaction between cancer cells and bone marrow-derived stromal cells implicated in tumor invasion, as majority of human gastric cancer is associated with loss or dysfunction of TGF-beta signaling [2]. It should be examined whether or not bone marrow-derived cells in gastric cancer microenvironment produce MMP and then encourage tumor cells to invade into the stroma. Moreover, gastric cancer originating from bone marrow-derived cells has been reported in mouse models [32]. However, Cogle et al. [33] reported that bone marrow stem cells mimic cancer but do not initiate it in human cancers.

Molecular Mechanisms of Gastric Carcinogenesis

Multiple genetic and epigenetic alterations in oncogenes, tumor suppressor genes, cell cycle regulators, cell adhesion molecules and DNA repair genes, as well as genetic instability and telomerase activation are responsible for tumorigenesis and progression of gastric cancer [1, 2, 31]. Differences exist in the pathways leading to intestinal and diffuse types of gastric carcinoma.

Inactivation of various genes including p16, hMHL1, CDH1, RAR-beta2, pS2 and RUNX3 by DNA methylation is involved in two distinct major genetic pathways of gastric cancer [2, 3, 34]. Hypermethylation of the p16 and of hMLH1 promoters is preferentially associated with intestinal type gastric carcinoma, whereas concordant hypermethylation of the CDH1 and RAR-beta2 promoters is predominantly detected in diffuse type gastric carcinoma. Loss or reduction of RUNX3 and pS2 expression by promoter methylation is a common event in both types of gastric carcinoma. In addition to promoter methylation, acetylated histone H4 is reduced in the majority of both types of gastric carcinoma [2]. Histone H4 is progressively deacetylated from the early stage to the late stage of the multi-step carcinogenesis of the stomach. Moreover, histone H3 acetylation is associated with reduced p21 (WAF1/CIP1) expression by gastric carcinoma [35].

Among these epigenetic events, RUNX3, a Runt domain transcription factor involved in TGF-beta signaling, is silenced in 63% gastric cancer as well as in 73% of hepatocellular carcinoma, 70% of bile duct cancer, 75% of pancreas cancer, 62% of laryngeal cancer, 46% of lung cancer, 25% of breast cancer and 23% of prostate cancer and 5% of colon cancer. Interestingly, inactivation of

RUNX 3 by promoter methylation is found in 8% of chronic gastritis, 28% of intestinal metaplasia, and 27% of gastric adenoma, but not in chronic hepatitis B [2]. These findings suggest that RUNX3 is a target for epigenetic gene silencing already at early stage of gastric carcinogenesis.

In the multi-step process of intestinal type gastric carcinoma, genetic instability and hyperplasia of hTERT positive stem cells precede replication error at the D1S191 locus, DNA hypermethylation at the D17S5 locus, pS2 loss, RAR-beta2 loss, RUNX3 loss, abnormal transcripts of CD44 and p53 mutation. All of these changes accumulate in at least 30% of incomplete intestinal metaplasia and are common events in intestinal type gastric cancer. An adenoma to carcinoma sequence is observed on around 20% of gastric adenoma with APC mutations. Molecular events associated with this sequence are loss of heterozygosity and mutation of p53, reduced p27 expression, loss of RUNX3, over-expression of cycline E and abnormal c-met transcription. The resulting advanced intestinal type gastric carcinomas frequently exhibit DCC loss, APC mutation, 1q LOH, loss of p27, reduced TGF beta-receptor expression, reduced nm23 and c-erbB2 gene amplification [1–3].

Meanwhile, diffuse type gastric carcinogenesis involves LOH at chromosome 17p, LOH or mutation of p53, LUNX3 loss and mutation or loss of E-cadherin. Gene amplification of K-sam, c-met and cycline E confers progression and metastasis. Several of the above molecular events may be present in mixed gastric cancer that have both intestinal and diffuse components [1–3].

Meta-analysis of epidemiological studies and animal models shows that both intestinal and diffuse types of

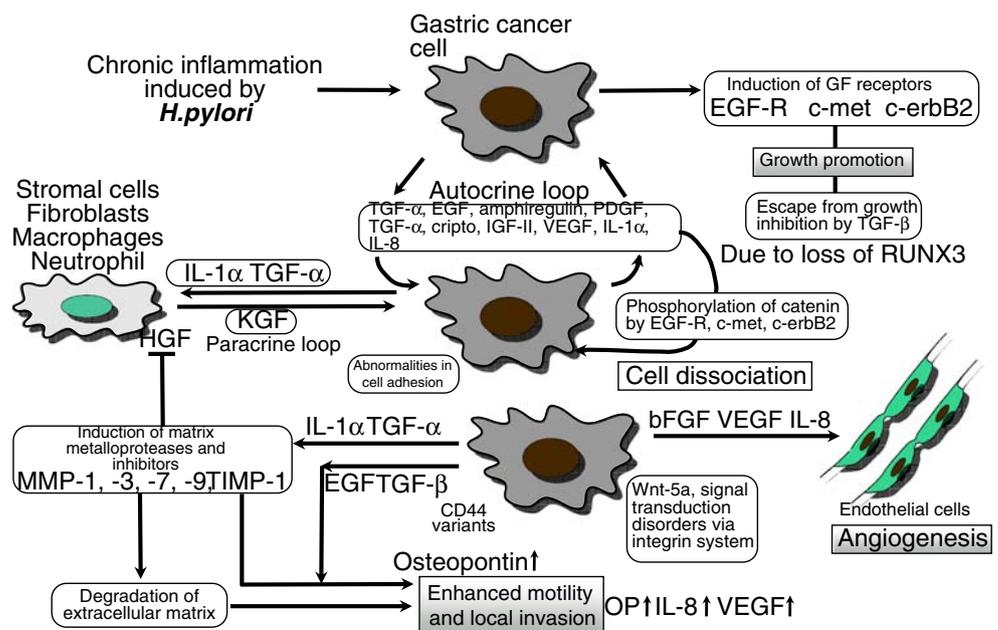
gastric cancer are equally associated with *H. pylori* infection [36, 37]. However, *H. pylori* infection may play a role only in the initial steps of gastric carcinogenesis. Importantly, younger *H. pylori*-infected patients have a higher relative risk for gastric cancer than older patients. In general, diffuse type gastric cancers occur mostly in younger patients. Hereditary diffuse gastric cancer caused by germ line E-cadherin mutations is also observed in younger age group [38]. We recently found that a low CagA IgG titer is a useful biomarker to identify a high-risk of gastric cancer and current smoking exhibits significantly higher risk for diffuse type than intestinal type gastric cancers, while radiation risk is significant only for nonsmokers and diffuse type gastric cancers [39]. Differences in *H. pylori* strain, patient age, exogenous and endogenous carcinogens and host genetic factors may be implicated in two distinct major genetic pathways for gastric carcinogenesis.

Abnormal Growth Factor/Cytokine Network in Gastric Cancer

Gastric cancer cells express a wide array of growth factors and cytokines that act via autocrine, paracrine and juxtacrine mechanisms in the tumor microenvironment [2, 3]. These complex interactions between tumor cells and stromal cells confer morphogenesis, angiogenesis, invasion and metastasis [Fig. 1]. Again the expression of these mediators varies depending on the histological subtype.

The EGF family including EGF, TGF alpha, cripto and amphiregulin (AR) are commonly overexpressed in intestinal

Fig. 1 Tumor microenvironment of gastric cancer is composed of interaction between cancer cells and stromal cells via growth factor/cytokine network



type carcinoma. Meanwhile TGF beta, insulin-like growth factor II (IGF II) and bFGF are predominantly overexpressed in the diffuse type carcinoma. Co-expression of EGF/TGF alpha and EGFR correlates well with the biological malignancy, as these factors induce metalloproteinase [3]. Overexpression of cripto is frequently associated with intestinal metaplasia and gastric adenoma [3]. Akagi et al. [40] reported that gastric cancer cells express neutrophilin-1 (NRP-1), a co-receptor for VEGF receptor 2 endothelial cells. EGF induces both NRP-1 and VEGF expression, suggesting that regulation of NRP-1 expression in gastric cancer is intimately associated with the EGF/EGFR system.

IL-1 alpha is produced not only by activated macrophages but also by gastric cancer cells. It acts as an autocrine growth factor for gastric carcinoma cells and plays a pivotal role as a trigger for the induction of EGF and EGF receptor [2, 3]. IL-6 also acts in an autocrine fashion to stimulate gastric cancer cells. IL-1 alpha and IL-6 both induces the expression of each other by tumor cells. Serum IL-6 in patients with gastric cancer is significantly correlated with tumor stage, depth of tumor invasion, lymphatic invasion, venous invasion and hepatic metastasis, suggesting that IL-6 may be useful for a prognostic factor of gastric cancer [41]. Interestingly, Lin et al. [42] recently found that IL-6 induces gastric cancer cell invasion via activation of the c-Src/RhoA/ROCK signaling pathway. Overexpression of these cytokines, IL-8 and growth factors by the tumor cells may be caused mainly by constitutive NF-kB activation due to constitutive activation of several upstream kinases [12].

More than 80% of primary gastric cancer express IL-8 and IL-8 receptor and this co-expression correlate directly with tumor vascularity and tumor invasion [43]. IL-8 enhances expression of EGF receptor, type IV collagenase (MMP9), VEGF and IL-8 mRNA itself by gastric cancer cells, while reducing E-cadherin mRNA expression [44]. Takehara et al. [45] recently reported that the COX-2 pathway might be also involved in IL-8 production in gastric cancer cells. In addition to IL-8, majority of gastric carcinoma express IL-11 and IL-11 receptor alpha and IL-11 promotes the migration of gastric cancer cells by the activation of the phosphatidylinositol-3 kinase pathway [46]. Moreover, recent studies show that expression of IL-12 and IL-18 also confers progression and metastasis [47, 48].

The negative growth factor TGF beta is frequently overexpressed in gastric carcinoma, particularly in diffuse type carcinoma with productive fibrosis [2]. Hawinkels et al. [49] reported that active TGF beta 1 is present in the tumor cells and fibroblasts, and high tumor TGF beta activity are significantly associated with worse survival of the patients. More importantly, recent *in vivo* and *in vitro* studies of Mishra group [50, 51] demonstrate that inactivation of ELF/TGF-beta signaling plays a key role in the

development of gastric carcinoma. Embryonic Liver Fodrin (ELF) is a beta-spectrin, an adaptor protein that plays an essential role in the propagation of TGF beta signaling. She found that loss of ELF and reduced Smad4 expression are observed in human gastric cancer.

Angiogenic factors such as VEGF, bFGF and IL-8 are produced by tumor cells and results neovascularisation within gastric carcinoma [2]. VEGF promote mainly angiogenesis and progression of intestinal type gastric cancer while bFGF has a stronger association with diffuse type gastric cancer. VEGF-C produced by tumor cells participates in the development of lymph node metastasis.

Stromal cells, especially fibroblasts stimulated by growth factors and cytokines such as IL-1 alpha, TGF alpha and TGF beta secrete hepatocyte growth factor/scatter factor (HGF/SF), which functions in a paracrine manner as a morphogen or motogen [2, 3]. HGF/SF from stromal cells binds to c-met on the tumor cells leading to enhanced progression via activation of c-met pathways. Moreover, keratinocyte growth factor (KGF) produced by gastric fibroblasts binds to KGF receptor, K-sam on tumor cells, resulting in the development of scirrhous type gastric cancer [52].

Osteopontin (OPN), also termed Eta-1 (early T-lymphocyte activation-1), which is a reported protein ligand of CD44, is overexpressed in 73% of gastric cancer [2, 3]. The co-expression of OPN and CD44 v9 in tumor cells correlates with the nodal metastasis in diffuse type gastric cancer. Wu et al. [53] recently reported that elevated plasma OPN level is significantly associated with gastric cancer development, invasive phenotype and survival, suggesting that plasma OPN might have potential usefulness as a diagnostic and prognostic factor for gastric cancer.

Regenerating islet-derived family, member 4 (Reg IV) is expressed in gastric cancer, colon cancer and pancreas cancer [54]. High Reg IV expression is associated with 5-FU resistance in gastric and colon cancer cells by inhibiting the mitochondrial apoptotic pathway, suggesting that expression of Reg IV is a marker for prediction of resistance to 5-FU chemotherapy in patients with gastric cancer [55]. Reg IV also induces phosphorylation of EGF receptor at Tyr992 and expression of Bcl2. Excitingly we found that Reg IV as well as MMP-10 are able to be useful for biomarkers to detect early stage of gastric cancer [56]. Moreover, Interferon inducible gene 6-16, GIP3 is also expressed in gastric cancers and inhibits mitochondrial-mediated apoptosis in gastric cancer cells, suggesting that 6-16 may be a new target for cancer therapy [57].

Wnt-5a is a representative of Wnt proteins that activate the beta-catenin-independent pathway. However, the functions of Wnt-5a in human cancer are controversial. Wnt-5a inhibits proliferation, migration, and invasiveness in thyroid tumor and colorectal cancer cell lines [58, 59], whereas

Wnt-5a expression is correlated with cell motility and invasiveness in melanoma cells and breast cancer cells with tumor-associated macrophages [60, 61]. Kurayoshi et al. [62] showed that expression of Wnt-5a is correlated with aggressiveness of gastric cancer by stimulating cell migration and invasion. Wnt-5a activates PKC and JNK, thereby leading to the activation of FAK and paxillin. Wnt-5a and extracellular matrix bind to Frizzled (Wnt-5a receptor) and integrin, and cooperatively activate a signaling cascade to stimulate cell migration. Wnt-5a may be a novel biomarker of prognostic factor and also a therapeutic target for gastric cancer [62].

These events of abnormal growth factor/cytokine networks in the tumor microenvironment will no doubt provide a great deal of information on new approaches for biomarkers and effective therapeutic targeting of gastric cancer.

Open Access This article is distributed under the terms of the Creative Commons Attribution Noncommercial License which permits any noncommercial use, distribution, and reproduction in any medium, provided the original author(s) and source are credited.

References

- Smith MG, Hold LG, Tahara E et al (2006) Cellular and molecular aspects of gastric cancer. *World J Gastroenterol* 12:2979–2990
- Tahara E (2004) Genetic pathways of two types of gastric cancer. In: Buffler P, Rice J, Bann R, Bird M, Boffeta P (eds) *Mechanisms of carcinogenesis: contributions of molecular epidemiology*. IARC Scientific Publications No.157, Lyon, pp 327–349
- Tahara E (2005) Growth factors and oncogenes in gastrointestinal cancers. In: Meyers RA (ed) *Encyclopedia of molecular cell biology and molecular medicine*, vol 6. 2nd edn. Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim, pp 1–31
- Alm RA, Ling LS, Moir DT et al (1999) Genomic-sequence comparison of two unrelated isolates of the human gastric pathogen, *Helicobacter pylori*. *Nature* 397:5552–5559
- Zarrilli R, Ricci V, Romano M et al (1999) Molecular response of gastric epithelial cells to *Helicobacter pylori* induced cell damage. *Cell Microbiol* 1:93–99
- Kuipers EJ, Perez-Perez G, Meuwissen et al (1995) *Helicobacter pylori* and atrophic gastritis: importance of the cagA status. *J Natl Cancer Inst* 87:1777–1780
- Prinz C, Schoniger M, Rad R et al (2001) Key importance of the *Helicobacter pylori* adherence factor blood group antigen binding adhesion during chronic inflammation. *Cancer Res* 61:1903–1909
- Hatakeyama M (2004) Oncogenic mechanisms of the *Helicobacter pylori* cagA protein. *Nat Rev, Cancer* 4:688–694
- Isomoto H, Mizuta Y, Takeshima F et al (2000) Implication of NF-kappaB in *Helicobacter pylori*-associated gastritis. *Am J Gastroenterol* 95:2768–2776
- Sharma SA, Tummuru MK, Blaser MJ et al (1998) Activation of IL-8 gene expression by *Helicobacter pylori* is regulated by transcription factor nuclear-kappa B in gastric epithelial cells. *J Immunol* 160:2401–2407
- Haz RA, Rieder G, Stolte M et al (1997) Pattern of adhesion molecule expression on vascular endothelium in *Helicobacter pylori*-associated antral gastritis. *Gastroenterology* 112:1908–1919
- Nakanishi C, Toi M (2005) Nuclear factor-kB inhibitors as sensitizers to anticancer drugs. *Nat Rev, Cancer* 5:297–309
- William CS, Smalley W, DuBois RN (1997) Aspirin and potential mechanisms for colorectal cancer prevention. *J Clin Invest* 100:1325–1329
- Tujii M, DuBois RN (1995) Alterations in cellular adhesion and apoptosis in epithelial cells overexpressing prostaglandin endoperoxide synthase 2. *Cell* 83:493–501
- Jaiswal M, LaRusso NF, Gores GJ (2001) Nitric oxide in gastrointestinal epithelial cell carcinogenesis: linking inflammation to oncogenesis. *Am J Physiol Gastrointest Liver Physiol* 281:626–634
- Goto T, Haruma K, Kitadai Y et al (1999) Enhanced expression of inducible nitric oxide synthase and nitrotyrosine in gastric mucosa of gastric cancer patients. *Clin Cancer Res* 3:1411–1415
- El-Omar EM, Carrington M, Chow WH et al (2000) Interleukin-1 polymorphisms associated with increased risk of gastric cancer. *Nature* 404:398–402
- El-Omar EM, Rabbin CS, Gammon MD et al (2003) Increased risk of noncardia gastric cancer associated with proinflammatory cytokine gene polymorphisms. *Gastroenterology* 124:1193–1201
- Hayashi T, Imai K, Kusunoki Y et al (2005) Radiation exposure affects immunogenetical risk of stomach cancer among atomic bomb survivors. Paper presented at the 96th Annual meeting of AACR, Anaheim, 16–20 April, 2005
- Ohyauchi M, Imatani A, Yonechi M et al (2005) The polymorphism interleukin 8–251 A/T influences the susceptibility of *Helicobacter pylori* related gastric diseases in the Japanese population. *Gut* 54:330–335
- Burkert J, Wright NA, Akison MR (2006) Stem cells and cancer: an intimate relationship. *J Pathol* 209:287–297
- Akasaka Y, Saikawa Y, Fujita K et al (2005) Expression of a candidate marker for progenitor cells, Musashi-1, in the proliferative regions of human antrum and its decreased expression in intestinal metaplasia. *Histopathology* 47:348–356
- Murata H, Tsuji S, Tsuji M et al (2007) *Helicobacter pylori* infection induces candidate stem cell marker Musashi-1 in the human gastric epithelium. *Dig Dis Sci Jun* 5
- Maekita T, Nakazawa K, Mihara M et al (2006) High levels of aberrant DNA methylation in *Helicobacter pylori*-infected gastric mucosa and its possible association with gastric cancer risk. *Clin Cancer Res* 12:989–995
- Beattie TL, Zhou W, Robinson MO et al (1998) Reconstitution of human telomerase activity in vitro. *Curr Biol* 8:177–180
- Sarin KY, Cheung P, Gilson D et al (2005) Conditional telomerase induction causes proliferation of hair follicle stem cells. *Nature* 436:1048–1052
- Kuniyasu H, Yasui W, Yokozaki H et al (2000) *Helicobacter pylori* infection and carcinogenesis of the stomach. *Langenbeck's Arch Surg* 385:69–74
- Tauchi T, Shin-Ya K, Sashida G et al (2003) Activity of a novel G-quadruplex–interactive, telomestatin (SOT-095), against human leukemia cells: involvement of ATM-dependent DNA damage response pathways. *Oncogene* 22:5338–5347
- Tahara H, Shin-Ya K, Seimiya H et al (2006) G-quadruplex stabilization by telomestatin induces TRF2 protein dissociation from telomeres and anaphase bridge formation accompanied by loss of the 3' telomeric overhang in cancer cells. *Oncogene* 25:1955–1966
- Kondo K, Oue N, Mitani Y et al (2005) Loss of heterozygosity and histone hypoacetylation of the PINX1 gene are associated with reduced expression in gastric carcinoma. *Oncogene* 24:157–164
- Kitamura Y, Kometani K, Hashida H et al (2007) SMAD4-deficient intestinal tumors recruit CCR1 myeloid cells that promote invasion. *Nat Genet* 39:467–475
- Houghton J, Stoicov C, Nomura et al (2004) Gastric cancer originating from bone marrow-derived cells. *Science* 306:1568–1571
- Cogle CR, Theise ND, Fu D et al (2007) Bone marrow contributes to epithelial cancers in mice and humans as developmental mimicry. *Stem Cells* 25:1881–1887

34. Tahara E, Lotan R (2005) RUNX3 and retinoic acid receptor beta DNA methylation as novel targets for gastric therapy. *Current Cancer Therapy Reviews* 1:139–144
35. Mitani Y, Oue N, Hamai Y et al (2005) Histone H3 acetylation is associated with reduced p21 WAF1/CIP1 expression by gastric carcinoma. *J Pathol* 205:65–73
36. Huang LQ, Sridhar S, Chen Y et al (1998) Meta-analysis of the relationship between *Helicobacter pylori* seropositivity and gastric cancer. *Gastroenterology* 114:1169–1179
37. Estick GD, Lim LL, Byles JE et al (1999) Association of *Helicobacter pylori* infection with gastric carcinoma: a meta-analysis. *Am J Gastroenterol* 94:2373–2379
38. Brooks-Wilson AR, Kaurah P, Suriano G et al (2004) Germline E-cadherin mutations in hereditary diffuse gastric cancer: assessment of 42 new families and review of genetic screening criteria. *J Med Genet* 41:508–517
39. Suzuki G, Cullings H, Fujiwara S et al (2007) Low-positive antibody titer against *Helicobacter pylori* cytotoxin-associated genes A (CagA) may predict future gastric cancer better than simple seropositivity against *H. pylori* CagA against *H. pylori*. *Cancer Epidemiol Biomark Prev* 16:1224–1228
40. Akagi et al (2003) Induction of neutrophilin-1 and vascular endothelial growth factor by epidermal growth factor in human gastric cancer cells. *Br J Cancer* 88:796–802
41. Ashizawa T, Okada R, Suzuki Y et al (2005) Clinical significance of interleukin-6 (IL-6) in the spread of gastric cancer: role of IL-6 a prognostic factor. *Gastric Cancer* 8:124–131
42. Lin MT, Lin BR, Change CC et al (2007) IL-6 induces AGS gastric cancer cell invasion via activation of the c-Src/RhoA/ROCK signaling pathway. *Int J Cancer* 120:2600–2608
43. Kitadai Y, Haruma K, Sumii K et al (1998) Expression of interleukin-8 correlates with vascularity in human gastric carcinoma. *Am J Pathol* 152:93–100
44. Kitadai Y, Haruma K, Mukaida N et al (2000) Regulation of disease-progression genes in human gastric carcinoma by interleukin-8. *Clin Cancer Res* 6:2735–2740
45. Takehara H, Iwamoto J, Mizokami Y et al (2006) Involvement of cyclooxygenase-2-prostaglandin E2 pathway in interleukin-8 production in gastric cancer cells. *Dig Dis Sci* 51:2188–2197
46. Nakayama T, Yoshizaki A, Izumida S et al (2007) Expression of interleukin-11 (IL-11) and IL-11 receptor alpha in human gastric carcinoma and IL-11 upregulates the invasive activity of human gastric carcinoma cells. *Int J Oncol* 30:825–833
47. Thong-Ngam D, Tangkivannich P, Lerknimitr R et al (2006) Diagnostic role of serum interleukin-18 in gastric cancer patients. *World J Gastroenterol* 12:4473–4477
48. Ye ZB, Ma T, Li H et al (2007) Expression and significance of intratumoral interleukin-12 and interleukin-18 in human gastric carcinoma. *World J Gastroenterol* 13:1747–1751
49. Hawinkels LJ, Verspaget HW, van Duijn W et al (2007) Tissue level, activation and cellular localization of TGF-beta1 and association with survival in gastric cancer patients. *Br J Cancer* 97:398–404
50. Katuri V, Tang Y, Marshal B et al (2005) Inactivation of ELF / TGF-beta signaling in human gastrointestinal cancer. *Oncogene* 24:8012–8024
51. Kim SS, Shetty K, Katuri V et al (2006) TGF-beta signaling pathway inactivation and cell cycle deregulation in the development of gastric cancer: role of the beta-spectrin, ELF. *Biochem Biophys Res Commun* 344:1216–1223
52. Nakazawa K, Yashiro M, Hirakawa K (2003) Keratinocyte growth factor produced by gastric fibroblasts specifically stimulates proliferation of cancer cells from scirrhous gastric carcinoma. *Cancer Res* 63:8848–8852
53. Wu CY, Wu MS, Chiang EP et al (2007) Elevated plasma osteopontin associated with gastric cancer development, invasion and survival. *Gut* 56:782–789
54. Oue N, Mitani Y, Phyu Phyu A et al (2005) Expression and localization of Reg IV in human neoplastic and non-neoplastic tissues: Reg IV expression is associated with intestinal and neuroendocrine differentiation in gastric carcinoma. *J Pathol* 207:185–198
55. Mitani Y, Oue N, Matsumura S et al (2007) Reg IV is serum biomarker for gastric cancer patients and predicts response to 5-fluorouacil-based chemotherapy. *Oncogene* 25:1–11
56. Tahara E (2007) A model for comprehensive prevention of gastric cancer based on novel biomarkers and longitudinal health database of atomic bomb survivors. Paper presented at the 4th International Conference on Tumor Microenvironment: Progression, Therapy and Prevention, Florence, March 6–10, 2007
57. Tahara E Jr, Tahara H, Kanno M et al (2005) GIP3, An interferon inducible gene 6–16, is expressed in gastric cancers and inhibits mitochondrial apoptosis in gastric cancer cell line TMK-1 cell. *Cancer Immunol Immunother* 54:729–740
58. Kremenevskaja N, von Wasielewski R, Rao AS et al (2005) Wnt-5a has tumor suppressor activity in thyroid carcinoma. *Oncogene* 24:2144–54
59. Dejmeek A, Safholm A, Sjolander A et al (2005) Wnt-5a protein expression in primary duks B colon caners identifies a subgroup of patients with good prognosis. *Cancer Res.* 65:3449–3499
60. Weraratna AT, Jiang Y, Hostetter G et al (2002) Wnt-5a signaling directly affects cell motility and invasion of metastatic melanoma. *Cancer Cell* 1:270–288
61. Rukrop T, Klemm F, Hageman T et al (2006) Wnt-5a signaling is critical for macrophage-induced invasion of breast cancer cell line. *Proc Natl Acad Sci USA* 103:5454–5459
62. Kurayoshi M, Oue N, Yamamoto H et al (2006) Expression of Wnt-5a is correlated with aggressiveness of gastric cancer by stimulating cell migration and invasion. *Cancer Res* 66:10439–10448